



P22042.ST25

SEQUENCE LISTING

<110> MIYAWAKI, Atsushi
NAGAI, Takeharu

<120> A Fluorescent Protein

<130> P22042

<140> US 10/086,738

<141> 2002-03-04

<150> PCT/JP05/98922001

<151> 2001-03-05

<160> 8

<170> PatentIn version 3.2

<210> 1

<211> 1278

<212> DNA

<213> Artificial

<220>

<223> Recombinant Fluorescent Protein

<400> 1

atgaagaggc gctggaagaa aaacttcatt gccgtcagcg ctgccaaccg gttcaagaag	60
atctccagct ccggggcact ggggtctgca ggctacaaca gccacaacgt ctatatcatg	120
gccgacaagc agaagaacgg catcaaggcc aacttcaaga tccgccacaa catcgaggac	180
ggcggcgtgc agctcgccga ccactaccag cagaacaccc ccatcggcga cggccccgtg	240
ctgctgcccg acaaccacta cctgagccac cagtccgccc tgagcaaaga cccaacgag	300
aagcgcgatc acatggtcct gctggagttc gtgaccgccg ccgggatcac tctcggcattg	360
gacgagctgt acaaggggtgg cagcgggtggc atggtgagca agggcgagga gctgttcacc	420
gggggtggtgc ccatcctggt cgagctggac ggcgacgtaa acggccacaa gttcagcgtg	480
tccggcgagg gcgagggcga tgccacctac ggcaagctga ccctgaagtt catctgcacc	540
accggcaagc tgcccgtgcc ctggcccacc ctctgacca ccttcggcta cggcctgaag	600

```

tgcttcgccc gctaccccga ccacatgaag cagcacgact tcttcaagtc cgccatgccc 660
gaaggctacg tccaggagcg caccatcttc ttcaaggacg acggcaacta caagacccgc 720
gccgaggtga agttcgaggg cgacaccctg gtgaaccgca tcgagctgaa gggcatcgac 780
ttcaaggagg acggcaacat cctggggcac aagctggagt acaacggtac cggggaccaa 840
ctgacagaag agcagattgc agagttcaaa gaagccttct cattattcga caaggatggg 900
gacggcacca tcaccacaaa ggaacttggc accgttatga ggtcgcttgg acaaaaccca 960
acggaagcag aattgcagga tatgatcaat gaagtcgatg ctgatggcaa tggaacgatt 1020
tactttcctg aatttcttac tatgatggct agaaaaatga aggacacaga cagcgaagag 1080
gaaatccgag aagcattccg tgtttttgac aaggatggga acggctacat cagcgctgct 1140
cagttacgtc acgtcatgac aaacctcggg gagaagttaa cagatgaaga agttgatgaa 1200
atgataaggg aagcagatat cgatggtgat ggccaagtaa actatgaaga gtttgtacaa 1260
atgatgacag caaagtaa 1278

```

```

<210> 2
<211> 1284
<212> DNA
<213> Artificial

```

```

<220>
<223> Recombinant Fluorescent Protein

```

```

<400> 2
atgaagaggc gctggaagaa aaacttcatt gccgtcagcg ctgccaaccg gttcaagaag 60
atctccagct ccggggcact ggggtctgca ggctacaaca gcgacaacgt ctatatcatg 120
gccgacaagc agaagaacgg catcaaggcc aacttcaaga tccgccacaa catcgaggac 180
ggcggcgtgc agctcgccga ccactaccag cagaacaccc ccatcggcga cggccccgtg 240
ctgctgcccc acaaccacta cctgagcttc cagtccgccc tgagcaaaga ccccaacgag 300
aagcgcgatc acatggtcct gctggagttc gtgaccgccg ccgggatcac tctcggcatg 360
gacgagctgt acaaggtcga cgggtggcagc ggtggcaccg gtgtgagcaa gggcgaggag 420

```

```

ctgttcaccg ggggtggtgcc catcctggtc gagctggacg gcgacgtaaa cggccacaag 480
ttcagcgtgt ccggcgaggg cgagggcgat gccacctacg gcaagctgac cctgaagctc 540
atctgcacca ccggcaagct gcccgtgccc tggcccaccc tcgtgaccac cttcggctac 600
ggcctgaagt gtttcgcccc ctacccccgac cacatgaagc agcacgactt cttcaagtcc 660
gccatgcccc aaggctacgt ccaggagcgc accatcttct tcaaggacga cggcaactac 720
aagacccgcg ccgaggtgaa gttcgagggc gacaccctgg tgaaccgcat cgagctgaag 780
ggcatcgact tcaaggagga cggcaacatc ctggggcaca agctggagta caacggtacc 840
gaccaactga cagaagagca gattgcagag ttcaaagaag ctttctcatt attcgacaag 900
gatggggacg gcaccatcac caciaaggaa cttggcaccg ttatgaggtc gcttggacaa 960
aaccaaacgg aagcagaatt gcaggatatg atcaatgaag tcgatgctga tggcaatgga 1020
acgatttact ttcctgaatt tcttactatg atggctagaa aaatgaagga cacagacagc 1080
gaagaggaaa tccgagaagc attccgtggt tttgacaagg atgggaacgg ctacatcagc 1140
gctgctcagt tacgtcacgt catgacaaac ctcggggaga agttaacaga tgaagaagtt 1200
gatgaaatga taagggaagc agatatcgat ggtgatggcc aagtaaacta tgaagagttt 1260
gtacaaatga tgacagcaaa gtaa 1284

```

```

<210> 3
<211> 1284
<212> DNA
<213> Artificial

```

```

<220>
<223> Recombinant Fluorescent Protein

```

```

<400> 3
atgaagaggc gctggaagaa aaacttcatt gccgtcagcg ctgccaaccc gttcaagaag 60
atctccagct ccggggcact ggggtctgca ggctacaaca gcgacaacgt ctatatcatg 120
gccgacaagc agaagaacgg catcaaggcc aacttcaaga tccgccacaa catcgaggac 180
ggcggcgtgc agctcgccga ccactaccag cagaacaccc ccatcggcga cggccccgtg 240

```

```

ctgctgccccg acaaccacta cctgagcttc cagtccgccc tgagcaaaga cccaacgag 300
aagcgcgatac acatggtcct gctggagttc gtgaccgccc ccgggatcac tctcggcatg 360
gacgagctgtg acaaggtcga cgggtggcagc ggtggcaccg gtgtgagcaa gggcgaggag 420
ctgttcaccg  ggggtggtgcc catcctggtc gagctggacg gcgacgtaaa cggccacaag 480
ttcagcgtgtg ccggcgaggg cgagggcgat gccacctacg gcaagctgac cctgaagctc 540
atctgcacca  ccggcaagct gcccgtgccc tggcccaccc tcgtgaccac cttcggctac 600
ggcctgaagt  gcttcgcccc ctaccccgac cacatgaagc agcacgactt cttcaagtcc 660
gccatgccccg aaggctacgt ccaggagcgc accatcttct tcaaggacga cggcaactac 720
aagaccgcgcg ccgaggtgaa gttcgagggc gacaccctgg tgaaccgcat cgagctgaag 780
ggcatcgact  tcaaggagga cggcaacatc ctgggggcaca agctggagta caacggtacc 840
gaccaactga  cagaagagca gattgcagag ttcaaagaag ctttctcatt attcgacaag 900
gatggggacg  gcaccatcac cacaaggaa cttggcaccg ttatgaggtc gcttggacaa 960
aaccaacgg  aagcagaatt gcaggatatg atcaatgaag tcgatgctga tggcaatgga 1020
acgatttact  ttctgaatt tcttactatg atggctagaa aaatgaagga cacagacagc 1080
gaagaggaaa  tccgagaagc attccgtgtt tttgacaagg atgggaacgg ctacatcagc 1140
gctgctcagt  tacgtcacgt catgacaaac ctcggggaga agttaacaga tgaagaagtt 1200
gatgaaatga  taagggaagc agatatcgat ggtgatggcc aagtaaacta tgaagagttt 1260
gtacaaatga  tgacagcaaa gtaa 1284

```

<210> 4

<211> 5

<212> PRT

<213> Artificial

<220>

<223> Linker Peptide

<400> 4

Gly Gly Ser Gly Gly

1 5

<210> 5
 <211> 9
 <212> PRT
 <213> Artificial

<220>
 <223> Linker Peptide

<400> 5

Val Asp Gly Gly Ser Gly Gly Thr Gly
 1 5

<210> 6
 <211> 425
 <212> PRT
 <213> Artificial

<220>
 <223> Recombinant fluorescent protein

<400> 6

Met Lys Arg Arg Trp Lys Lys Asn Phe Ile Ala Val Ser Ala Ala Asn
 1 5 10 15

Arg Phe Lys Lys Ile Ser Ser Ser Gly Ala Leu Gly Ser Ala Gly Thr
 20 25 30

Asn Ser His Asn Val Tyr Ile Met Ala Asp Lys Gln Lys Asn Gly Ile
 35 40 45

Lys Ala Asn Phe Lys Ile Arg His Asn Ile Glu Asp Gly Gly Val Gln
 50 55 60

Leu Ala Asp His Tyr Gln Gln Asn Thr Pro Ile Gly Asp Gly Pro Val
 65 70 75 80

Leu Leu Pro Asp Asn His Tyr Leu Ser His Gln Ser Ala Leu Ser Lys

85

90

95

Asp Pro Asn Glu Lys Arg Asp His Met Val Leu Leu Glu Phe Val Thr
 100 105 110

Ala Ala Gly Ile Thr Leu Gly Met Asp Glu Leu Tyr Lys Gly Gly Ser
 115 120 125

Gly Gly Met Val Ser Lys Gly Glu Glu Leu Phe Thr Gly Val Val Pro
 130 135 140

Ile Leu Val Glu Leu Asp Gly Asp Val Asn Gly His Lys Phe Ser Val
 145 150 155 160

Ser Gly Glu Gly Glu Gly Asp Ala Thr Tyr Gly Lys Leu Thr Leu Lys
 165 170 175

Phe Ile Cys Thr Thr Gly Lys Leu Pro Val Pro Trp Pro Thr Leu Val
 180 185 190

Thr Thr Phe Gly Tyr Gly Leu Lys Cys Phe Ala Arg Tyr Pro Asp His
 195 200 205

Met Lys Gln His Asp Phe Phe Lys Ser Ala Met Pro Glu Gly Tyr Val
 210 215 220

Gln Glu Arg Thr Ile Phe Phe Lys Asp Asp Gly Asn Tyr Lys Thr Arg
 225 230 235 240

Ala Glu Val Lys Phe Glu Gly Asp Thr Leu Val Asn Arg Ile Glu Leu
 245 250 255

Lys Gly Ile Asp Phe Lys Glu Asp Gly Asn Ile Leu Gly His Lys Leu
 260 265 270

Glu Tyr Asn Gly Thr Gly Asp Gln Leu Thr Glu Glu Gln Ile Ala Glu

275

280

285

Phe Lys Glu Ala Phe Ser Leu Phe Asp Lys Asp Gly Asp Gly Thr Ile
 290 295 300

Thr Thr Lys Glu Leu Gly Thr Val Met Arg Ser Leu Gly Gln Asn Pro
 305 310 315 320

Thr Glu Ala Glu Leu Gln Asp Met Ile Asn Glu Val Asp Ala Asp Gly
 325 330 335

Asn Gly Thr Ile Tyr Phe Pro Glu Phe Leu Thr Met Met Ala Arg Lys
 340 345 350

Met Lys Asp Thr Asp Ser Glu Glu Glu Ile Arg Glu Ala Phe Arg Val
 355 360 365

Phe Asp Lys Asp Gly Asn Gly Tyr Ile Ser Ala Ala Gln Leu Arg His
 370 375 380

Val Met Thr Asn Leu Gly Glu Lys Leu Thr Asp Glu Glu Val Asp Glu
 385 390 395 400

Met Ile Arg Glu Ala Asp Ile Asp Gly Asp Gly Gln Val Asn Tyr Glu
 405 410 415

Glu Phe Val Gln Met Met Thr Ala Lys
 420 425

<210> 7
 <211> 427
 <212> PRT
 <213> Artificial

<220>
 <223> Recombinant fluorescent protein

<400> 7

Met Lys Arg Arg Trp Lys Lys Asn Phe Ile Ala Val Ser Ala Ala Asn
 1 5 10 15

Arg Phe Lys Lys Ile Ser Ser Ser Gly Ala Leu Gly Ser Ala Gly Tyr
 20 25 30

Asn Ser Asp Asn Val Tyr Ile Met Ala Asp Lys Gln Lys Asn Gly Ile
 35 40 45

Lys Ala Asn Phe Lys Ile Arg His Asn Ile Glu Asp Gly Gly Val Gln
 50 55 60

Leu Ala Asp His Tyr Gln Gln Asn Thr Pro Ile Gly Asp Gly Pro Val
 65 70 75 80

Leu Leu Pro Asp Asn His Tyr Leu Ser Phe Gln Ser Ala Leu Ser Lys
 85 90 95

Asp Pro Asn Glu Lys Arg Asp His Met Val Leu Leu Glu Phe Val Thr
 100 105 110

Ala Ala Gly Ile Thr Leu Gly Met Asp Glu Leu Tyr Lys Val Asp Gly
 115 120 125

Gly Ser Gly Gly Thr Gly Val Ser Lys Gly Glu Glu Leu Phe Thr Gly
 130 135 140

Val Val Pro Ile Leu Val Glu Leu Asp Gly Asp Val Asn Gly His Lys
 145 150 155 160

Phe Ser Val Ser Gly Glu Gly Glu Gly Asp Ala Thr Tyr Gly Lys Leu
 165 170 175

Thr Leu Lys Leu Ile Cys Thr Thr Gly Lys Leu Pro Val Pro Trp Pro
 180 185 190

Thr Leu Val Thr Thr Phe Gly Tyr Gly Leu Lys Cys Phe Ala Arg Tyr
 195 200 205

Pro Asp His Met Lys Gln His Asp Phe Phe Lys Ser Ala Met Pro Glu
 210 215 220

Gly Tyr Val Gln Glu Arg Thr Ile Phe Phe Lys Asp Asp Gly Asn Tyr
 225 230 235 240

Lys Thr Arg Ala Glu Val Lys Phe Glu Gly Asp Thr Leu Val Asn Arg
 245 250 255

Ile Glu Leu Lys Gly Ile Asp Phe Lys Glu Asp Gly Asn Ile Leu Gly
 260 265 270

His Lys Leu Glu Tyr Asn Gly Thr Asp Gln Leu Thr Glu Glu Gln Ile
 275 280 285

Ala Glu Phe Lys Glu Ala Phe Ser Leu Phe Asp Lys Asp Gly Asp Gly
 290 295 300

Thr Ile Thr Thr Lys Glu Leu Gly Thr Val Met Arg Ser Leu Gly Gln
 305 310 315 320

Asn Pro Thr Glu Ala Glu Leu Gln Asp Met Ile Asn Glu Val Asp Ala
 325 330 335

Asp Gly Asn Gly Thr Ile Tyr Phe Pro Glu Phe Leu Thr Met Met Ala
 340 345 350

Arg Lys Met Lys Asp Thr Asp Ser Glu Glu Glu Ile Arg Glu Ala Phe
 355 360 365

Arg Val Phe Asp Lys Asp Gly Asn Gly Tyr Ile Ser Ala Ala Gln Leu
 370 375 380

Arg His Val Met Thr Asn Leu Gly Glu Lys Leu Thr Asp Glu Glu Val
 385 390 395 400

Asp Glu Met Ile Arg Glu Ala Asp Ile Asp Gly Asp Gly Gln Val Asn
 405 410 415

Tyr Glu Glu Phe Val Gln Met Met Thr Ala Lys
 420 425

<210> 8

<211> 427

<212> PRT

<213> Artificial

<220>

<223> Recombinant fluorescent protein

<400> 8

Met Lys Arg Arg Trp Lys Lys Asn Phe Ile Ala Val Ser Ala Ala Asn
 1 5 10 15

Arg Phe Lys Lys Ile Ser Ser Ser Gly Ala Leu Gly Ser Ala Gly Tyr
 20 25 30

Asn Ser Asp Asn Val Tyr Ile Met Ala Asp Lys Gln Lys Asn Gly Ile
 35 40 45

Lys Ala Asn Phe Lys Ile Arg His Asn Ile Glu Asp Gly Gly Val Gln
 50 55 60

Leu Ala Asp His Tyr Gln Gln Asn Thr Pro Ile Gly Asp Gly Pro Val
 65 70 75 80

Leu Leu Pro Asp Asn His Tyr Leu Ser Phe Gln Ser Ala Leu Ser Lys
 85 90 95

Asp Pro Asn Glu Lys Arg Asp His Met Val Leu Leu Glu Phe Val Thr

100	105	110
Ala Ala Gly Ile Thr Leu Gly Met Asp Glu Leu Tyr Lys Val Asp Gly		
115	120	125
Gly Ser Gly Gly Thr Gly Val Ser Lys Gly Glu Glu Leu Phe Thr Gly		
130	135	140
Val Val Pro Ile Leu Val Glu Leu Asp Gly Asp Val Asn Gly His Lys		
145	150	155
Phe Ser Val Ser Gly Glu Gly Glu Gly Asp Ala Thr Tyr Gly Lys Leu		
165	170	175
Thr Leu Lys Leu Ile Cys Thr Thr Gly Lys Leu Pro Val Pro Trp Pro		
180	185	190
Thr Leu Val Thr Thr Phe Gly Tyr Gly Leu Lys Cys Phe Ala Arg Tyr		
195	200	205
Pro Asp His Met Lys Gln His Asp Phe Phe Lys Ser Ala Met Pro Glu		
210	215	220
Gly Tyr Val Gln Glu Arg Thr Ile Phe Phe Lys Asp Asp Gly Asn Tyr		
225	230	235
Lys Thr Arg Ala Glu Val Lys Phe Glu Gly Asp Thr Leu Val Asn Arg		
245	250	255
Ile Glu Leu Lys Gly Ile Asp Phe Lys Glu Asp Gly Asn Ile Leu Gly		
260	265	270
His Lys Leu Glu Tyr Asn Gly Thr Asp Gln Leu Thr Glu Glu Gln Ile		
275	280	285
Ala Glu Phe Lys Glu Ala Phe Ser Leu Phe Asp Lys Asp Gly Asp Gly		

290

295

300

Thr Ile Thr Thr Lys Glu Leu Gly Thr Val Met Arg Ser Leu Gly Gln
 305 310 315 320

Asn Pro Thr Glu Ala Glu Leu Gln Asp Met Ile Asn Glu Val Asp Ala
 325 330 335

Asp Gly Asn Gly Thr Ile Tyr Phe Pro Glu Phe Leu Thr Met Met Ala
 340 345 350

Arg Lys Met Lys Asp Thr Asp Ser Glu Glu Glu Ile Arg Glu Ala Phe
 355 360 365

Arg Val Phe Asp Lys Asp Gly Asn Gly Tyr Ile Ser Ala Ala Gln Leu
 370 375 380

Arg His Val Met Thr Asn Leu Gly Glu Lys Leu Thr Asp Glu Glu Val
 385 390 395 400

Asp Glu Met Ile Arg Glu Ala Asp Ile Asp Gly Asp Gly Gln Val Asn
 405 410 415

Tyr Glu Glu Phe Val Gln Met Met Thr Ala Lys
 420 425